### PERSOONIA

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### ARMILLARIA STAUDE IN THE CAMEROON REPUBLIC

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The intricacies of the nomenclature of the African Armillaria variously known as Clitocybe elegans, A. heimii, and A. fuscipes, and its relationship to A. mellea var. camerunensis are discussed. A full illustrated description of recently collected material from the Guinea-Congo rainforest is given, including details of the wall-structure of the basidiospores.

### **PREAMBLE**

The Korup rainforest reserve is situated in the Western Province of the Republic of Cameroon. In parallel to other large tracts of dense natural rainforest in the tropics the fungus flora is rich and diverse. There is a vast array of saprotrophs and several ectomycorrhizal basidiomycetes, the latter a rather unexpected group of fungi, although such a biotrophic state is rather more widely distributed in the tropics than previously thought. The saprotrophs include a wide range of taxa. Some such as the marasmioid agarics grow on leaves and twigs, some especially the corticiaceous fungi and centrally stipitate polypores, especially members of the genus *Microporus*, grow on small branches; and others utilise rather larger segments of wood, such as *Lentinus* and numerous polypores. Members of the genus *Mycena* are comparatively uncommon, although there are some widespread species, and their range of habitats is much the same as those species discussed by R. A. Maas Geesteranus in his series of authoritative articles on the temperate species.

Before Maas Geesteranus took up his present study he was recognised as the authority on the toothed fungi and as such prepared the account of species from the Congo region. In this work he recognised and illustrated *Donkia sanguinea* (Beeli) Maas G. later to be transferred to *Climacodon*. This fungus is extremely common in Korup especially after the first rains, and grows on small twigs as well as in larger troops on rather more substantial woody debris. It grows very rapidly and from small match-sized primordia in 7 days it reaches over 50 mm in diameter. It is in similar habitats that *Armillaria fuscipes* Petch grows, the subject of this paper which is dedicated to a mycologist who has contributed so much to our understanding of both temperate and tropical fungi.

## INTRODUCTION

During the division of Africa by the European imperialists in the nineteenth century those African kingdoms bordering the Gulf of Guinea were divided between France, Germany, and

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Great Britain. In 1884 Germany made the Cameroon a protectorate but after the First World War five sixths of this territory became part of French Equatorial Africa, a colony now dispersed amongst several autonomous countries. The rest, which was close to the Nigerian border, came under British rule. The first two of the European countries involved in this history have played an important role in the understanding of the fungal flora of the area; it is surprising that Great Britain with its studies in fungi of Sierra Leone, Ghana, Nigeria, etc. paid no attention to Cameroon. Cameroon borders in the southeast with Congo, whose rainforests spread into what is now Zaire, and after the First World War the attention of Belgian mycologists was drawn to the western part of this domain.

One of the German plant collectors active in Cameroon was Georg A. Zenker (1855–1922) celebrated in several genera of flowering plants including *Neozenkerina* Mildbrand, 1921. Although primarily collecting seed-plants, several specimens of the larger fungi he found especially around Bipinde in the southwest of the country were dispatched for examination to Paul Hennings then in Berlin. The results of this examination were published in four papers in Engler's Botanische Jahrbücher (Hennings, 1895, 1897, 1901, 1905). Unfortunately many of Hennings' specimens were lost in the air raids on Berlin during World War II but in the Royal Botanic Garden, Edinburgh (E) duplicates of many of Zenker's collections are maintained. One of the taxa described by Hennings (1895) was *Armillaria mellea* var. *camerunensis* and a collection is housed in E, viz. *Zenker 2165*. Hennings' fungus is actually based on a Dusén collection ('n. 1a - 19 Juli 1892') from near Bomana ('c. 670 m an faulenden Baumstämmen rasig'); Bomana is located between Buea and Douala in southern Cameroon. In contrast Zenker's collection eight years later was from Bipinde, then an area of virgin forest southeast of Douala.

Although Bresadola (1890) had earlier published the results of an examination of collections made by Joanne Braun, little was added to our knowledge of Cameroon fungi until the closing years of the Second World War. It was at this stage that the French mycologists paid particular attention to the resources of their African colonies, adding to the already long tradition of tropical mycology, of which Narcisse Patouillard was a leading figure. Thus Heim, based in Paris, studied a species of Armillaria (as Clitocybe (Armillariella)) from tropical Africa and the similarities between his collections and recent ones from rainforest in the Western Province of Cameroon (Korup) are the subject of this paper.

### THE KORUP ARMILLARIA

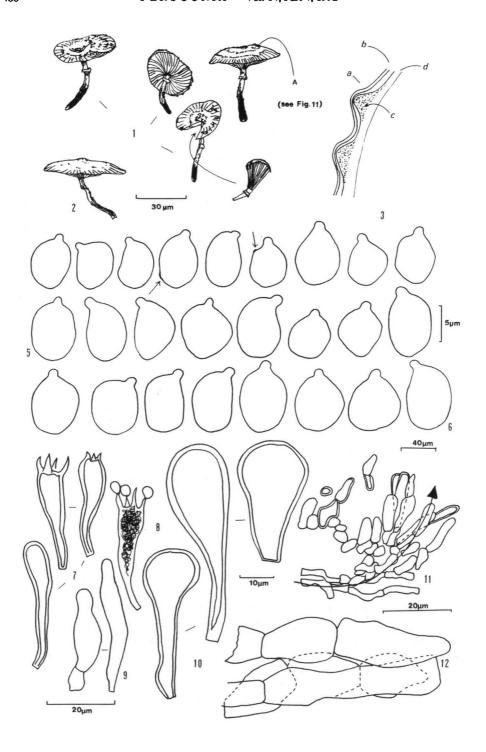
Pileus 12-42 mm, plano-convex soon plane or slightly depressed at centre, pale brownish buff, very pale to white at margin, dry, ornamented with very tiny dark brown scales, denser towards disc but soon lost by weathering. Gills whitish, tinged pinkish buff with age, adnexo-adnate with slight but not prominent decurrent tooth. Stipe  $20-30 \times 2.5-4$  mm (> 5 mm at base) slender, cylindrical, darkening, roughened with white fibrils above the ring, with small white scales on a dark background up to a cortinate ring. Veil annulate, collapsing, whitish with a few, small marginal black smudges (not as pronounced as in *Armillaria ostoyae*). Flesh thin in pileus, pinkish buff, more fibrous in stipe, similarly coloured at apex, brownish downwards to base; smell not distinct; taste not particularly unpleasant. Spore-print creamcolour (C).

Basidiospores  $6-8 \times 4.8-5.2(-5.7)$  µm, ellipsoid-ovoid, slightly angular in some views, others almost trapezoid and with irregular broad, blunt angles, smooth or in some with slight irregularities in the outline, slightly thick-walled, with no apical differentiation, with pronounced, sometimes even snout-like, hilar appendage (Figs. 4, 5, 15, 16). Basidia 33-37 × 6.5-10 μm (excl. sterigmata 2.2-3.3 μm long), 4-spored, cylindric slightly tapered downwards, hyaline, thin-walled often containing a large amount of honey-coloured, colloidal material (Fig. 8); crassobasidia 33-40 µm long, 0-4-spored, thick-walled, yellowish, in ammoniacal solutions, very sparse, possibly more frequent in older basidiomes (Fig. 7). Pleurocystidia elongate-cylindric, 10.9–19.5 × 8.7–15.3 μm with pedicel 22 μm long, resembling basidioles intermixed with deeply seated, thick-walled, yellow cells with rounded heads resembling a chlamydospore on a long pedicel 33-44 × 11 μm, or clavate, 33-39 × 8.7-9.8 μm, to subcapitate scattered or somewhat clustered where gill thickens at branches or at veins (Fig. 10); cheilocystidia resembling basidioles or slightly differentiated at apex to become subcapitate, smooth, hyaline, thin-walled, 23-30 × 4-5 µm, or clavate to cylindric with obtuse apex, some with colloidal material and intermixed with fertile basidia (Fig. 9). Hymenophoral trama bilateral with a honey coloured mediostratum of non-gelatinized hyphae and divergent lateral strata supporting a narrow subhymenium, lacking laticiferous hyphae and more open; floccose towards the gill-base, consisting of hyphae 3-4.4 µm broad, forming a band > 66 µm, mediostratum expanding to 175-218 µm broad with age composed of hyphae 11-22 µm broad, many swollen to ellipsoid almost obliterating lateral strata which become slightly gelatinized and intertwined with the subhymenium. Pileipellis (Figs. 11, 12) consisting of a disrupted outer layer (mediopellis) of cylindric, ellipsoid and/or shortened cells, forming a basal layer to aggregations of similar, brown or hyaline generally smooth-walled, ellipsoid, cylindric, utriform cells of suprapellis,  $(13-)50-75 \times 11-30 \mu m$ , intermixed with more filamentous units (4.4 µm broad) which form erect to suberect scales often recurved at their apex, with many elements disarticulating; mediopellis passing into a hyaline, disrupted open layer with little demarcation (except for slight honey colouring) from the open, hyaline context, composed of contorted, shortened elements which become more organized towards the hymenium where it takes on the form of cylindric hyphae, joining the mediostratum. Clampconnections apparently absent.

In cluster on fallen tree, in dense shade of fairly young growth overshadowed by grove of towering legumes, Korup Rainforest Reserve: Transect P, 25.III.1989, *Wat.* 22261: Edge of trail from Field 67 to Ekunda-Kunda, 11.IV.1990, *Wat.* 22262 (Figs. 1, 2).

In older specimens the brown disarticulating cells of the suprapellis are only located as a residual covering in radial depressions of the pileus, the pileipellis then becomes a weathered layer. Indeed in *Wat.* 22262 the scales are almost lost completely probably because of the appalling weather conditions.

Both collections were from a natural area of lowland rainforest lying between the Akva-Korup and Ndian rivers in southwest Cameroon and within the Guinea-Congonian refuge area into which the forest was reduced during the Pleistocene (Gartlan, 1974). It is characterized by both an assemblage of legumes in subfamily Caesalpinioideae, tribes Amhersticae and Detaricae on freely draining, acidic, sandy soil with a mean annual rainfall of c. 5500 mm, having a distinct dry season from December to February. The mean monthly temperature is in the range 24–30°C. One collection of A. fuscipes was made towards the end of the dry season, the other in the early part of the rainy season. The rainforest in which they occurred is at low elevation (c. 1000 m) and consists of an estimated 700 different arborescent taxa within the Korup forest alone. 411 taxa of flowering plants within defined subplots have been recog-



nized amongst all living stems > 30 cm girth at breast height (1.3 m). Unfortunately neither of the *Armillaria* collections could be tied to a specific identifiable species nor one of the 447 so far unidentifiable. The first collection occurred in an  $80 \times 80$  m sample plot dominated by *Microbelinia bisulcata* A. Chev. (15 m² ha-¹ area) with accompanying *Tetrabelinia moreliana* Aubr. (5 m² ha-¹ basal area) and a little *T. bifoliolata* (Harms) Hartman – all three ectotrophic Leguminosae.

The Cameroon material agrees with the description of Clitocybe elegans Heim, Armillaria heimii Pegler, and A. fuscipes Petch and it differs from A. mellea and its allies in the smaller number of individuals in a cluster, the slender more delicate growth, the pale almost white pileus especially towards the margin, the low density of pileus-scales and floccose stipes lacking a membranous, ample ring.

The basidiospores of the February collection have been studied by both light and electron microscopy. By using light microscopy, including fluorescent and phase contrast techniques, with or without staining, the spores appear apparently smooth or at most only slightly irregular. In contrast electron-microscope studies show a low contoured ornamentation which is poorly defined with the scanning electron microscope but S. Helfer, who has undertaken the sectioning and transmission electron microscope studies, has demonstrated the same complexity of wall structure now seen in a whole series of Armillaria species. The wall is at least three-layered with a thin outermost layer, a broad medial layer in which lenses of electron dense material are embedded (Fig. 3), and a lower basal layer. The lenses are located beneath the ridges as shown for A. mellea (Vahl: Fr.) Kummer, A. hinnulea Kile & Watling etc. (Bennell & al., 1985).

### EARLIER STUDIES

## (a) Clitocybe elegans Heim

Heim & Bouriquet as long ago as 1937 had studied a species of Armillaria (as Clitocybe) amongst the possible causal agents of a disease of Albizzia (Leguminosae) in Madagascar, grown there for wood-production. Indeed it was during this study that the name Clitocybe elegans Heim was first introduced, although it was over twenty-five years later that the epithet was validated (Heim, 1963). Fortunately, however, the Latin diagnosis was accompanied by both additional field data and a coloured plate. A full description was given of the basidiomes from a rotten trunk amongst talus in northern Madagascar (26.I.1935; Montagne d'Ambre) and not from the collections on Albizzia. The former material (Heim 264) which has been examined is in liquid preservative and is cited as the type; it is figured with other collections in a coloured plate (Heim, 1963; Kile & Watling, 1988). This species is also recorded from coastal forests near Douala (Batanga), Cameroon and from summit forest

Figs. 1-5,7-12. Armillaria fuscipes. Fig. 6. A. mellea var. camerunensis. — 1. Habit sketch (Wat. 22261). — 2. Habit sketch (Wat. 22262). — 3. Diagram of basidiospore-wall showing lenses of electron-dense material. — 4-6. Basidiospores (4, 7-12 Wat.. 22261; 5. Wat 22262; 6. Zenker 2165). — 7. Three crassobasidia. — 8. Basidium with colloidal contents. — 9. Two cheilocystidia. — 10. Three thick-walled pleurocystidia. — 11. Section of pileipellis showing orientation of scale; see A in Fig. 1. — 12. Elements of scale.

(1000 m: Mont Tonkom) in the Ivory Coast; the Batanga collection is also figured by Heim (1963).

In Madagascar Clitocybe elegans also attacks coffee (Coffea – Rubiaceae) and Dadant (1963) has made a study of this and the relationships between C. elegans and Trichoderma viride Pers. He successfully cultured his isolates and these basidiomes so produced agreed with E. African material grown in culture in Britain from material supplied by M. Ivory (pers. comm.). Later, Heim (1967) recorded the same species from New Guinea and from the Central African Republic.

All workers have recognised the close affinities of *Clitocybe elegans* to *Armillaria mellea* (= *Armillariella* P. Karst. s. Singer), the type of the genus *Armillaria* but because of their circumscription of genera within the white-spored tricholomataceous agarics the French mycologists did not transfer the taxon to *Armillaria* (see Watling & al., 1982).

Pegler (1977) realised that *Clitocybe elegans* unfortunately would be pre-dated if transferred to *Armillaria* as Beeli (1927) had already described *Armillaria elegans* from Diobo Akuba, Congo; Thoen (1969) later showed this to be a member of the genus *Cystoderma*. Pegler (1977) coined the epithet *heimii* for the African fungus to recognise its autonomy and Heim's contribution not only to the study of this particular taxon but agaricology as a whole, in Africa, the tropics and elsewhere.

## (b) Armillaria mellea var. camerunensis Henn.

As indicated in the introduction Hennings (1895) described a variety of A. mellea based on a collection from Cameroon by Dusén. Zenker made a later collection in 1900 which Hennings labelled 'A. camerunensis' thus raising the variety to specific rank. However it was not until later that Singer (1986) officially made the transfer to Armillariella, which is synonymous with Armillaria as here understood. Hennings' manuscript name is not valid according to the Rules of Botanical Nomenclature. The original description of A. mellea var. camerunensis is offered as follows:

'A. mellea Vahl in Fl. Dan. t. 103 var. camerunensis P. Henn.; pileo carnosulo, convexo-explanato, ½ -1 cm diametro, rufo-brunneo, dense granulato- vel verrucoso-squamoso, squamis parvis atris subconicis vel depressis, margine primo involuto dein explanato, substriato; stipite farcto, 1-2 cm longo, 2-3 mm crasso, subsquamoso, substriato, laete brunneo, parte superiore annulo amplo, membranaceo-floccoso, albo, patente; lamellis sinuoso-adnatis, vix decurrentibus, subconfertis, pallidis; sporis subglobosis, levibus, hyalinis, 7-8 m, basidiis clavatis.'

'Kamerun, bei Bomana, c. 670 m, an faulenden Baumstämmen rasig (P. Dusén n. 1<sup>a</sup>-19 Juli 1892).'

'Eine sehr zierliche und kleine Form, die äußerlich der typischen Art sehr ähnlich, aber durch die nicht herablaufenden Lamellen etwas verschieden ist. Bei N'dian wurden von Herrn Dusén unter n. 39 a lange Rhizomorphenstränge an faulenden Stammen gesammelt, die von derartigen Mycelien des Pilzes nicht verschieden sind und wahrscheinlich zu demselben gehören.'

Armillaria mellea var. camerunensis has only otherwise been recorded by Beeli (1927) along with three other species of Armillaria from the Congo, viz. A. elegans Beeli, A. dacty-liophora (Lév.) Beeli, and A. pelliculata Beeli. Beeli's collection of var. camerunensis was from soil in an inundated forest at Eala, Armillaria dactyliophora (Lév.) Beeli is Lentinus sajor-caju (Fr.) Fr., a widespread, edible member of the genus, of which L. dactyliophora Lév. is a synonym; A. elegans as indicated earlier is a member of the genus Cystoderma

(Thoen, 1969). Armillaria pelliculata was reported as not possessing basidiospores and its identity will remain a mystery until an agaric agreeing with the protologue is refound in Central Africa. The presence of a grey, pellicular, viscid (?) veil and infundibuliform pileus does not immediately indicate a species of Armillaria s. stricto (= Armillariella P. Karst. s. Singer 1951 et subseq.). It could be an Oudemansiella of which there are several morphotypes in the Guinea-Congo rainforests.

Examination of the material of A. camerunensis housed in E shows that there are differences between it and the Korup specimens. The material consists of sixteen annulate basidiomes, all quite small and relatively delicate. The stipes are longer than those found in the Korup collections and the diameter of the pileus is smaller. Although now quite mouldy the general facies of the Bipinde collections is different to that of the present specimens but agrees fairly well with Hennings' protologue. The basidiospores are also more in keeping with Hennings' description being  $6-8.3(-9.2) \times (4.5-)5.2-7 \,\mu m$  in the Bipinde material ('subglobosis') as opposed to  $6-8 \times 4.8-5.2(-5.7) \mu m$  (Figs. 4-6). Indeed the Korup collections agree in their spore-measurements with those for the type of A. fuscipes given by Chandra & Watling (1982) and for A. heimii given by Pegler (1977). A later range given by Pegler (1986) for A. fuscipes would cover the spore-ranges for the Bipinde and Korup specimens and there is no doubt the outline of the basidiospores of the two is close (Figs. 4-6). Unfortunately a further complication has been found in that when Singer (1986) made the new combination he wrote 'with the spores longitudinally ridged under the light microscope'. Although under the electron microscope differentiation of the basidiospore wall can be demonstrated it is only in one taxon, viz. A. duplicata (Berk.) Sacc., that the ornament is strong enough to form ridges (Chandra & Watling, 1982) visible under the light microscope. It has neither been seen in the Korup collections, and what is more significant nor in the Bipinde material (Zenker 2165); this may mean that Hennings had a broad concept of his taxon or has misidentified the later specimens from Cameroon.

In stature and general facies the Korup collections, which are uniform, differ from the material from Bipinde and it is proposed to maintain the two separately until a future successful search is made for *Armillariella camerunensis* (Henn.) Singer, and the taxon is redefined and cultured.

## (c) Armillaria fuscipes Petch

Petch (1909) described the species Armillaria fuscipes growing on the roots of Acacia decurrens (Wendl.) Willd. (Leguminosae), a plant which is extensively grown as a windbreak on tea plantations in Sri Lanka, and indicated that this new taxon killed the roots. Petch reiterated this, both in a book on diseases of the tea-bush (Petch, 1923) and in an account of the root diseases of trees (Petch, 1928). Although it has been considered by some to be A. mellea, Petch (1928) maintained A. fuscipes as an autonomous species.

Chandra & Watling (1982) have examined authentic and type material of A. fuscipes housed in the Royal Botanic Gardens, Kew (K) and in New Zealand (DDP - No. 35405) apparently originally from Peradeniya (Taylor, pers. comm.). They confirmed Petch's conviction that this is a distinct taxon but deferred from synonymizing Clitocybe elegans and A. fuscipes because of rather tantalizing small differences in rhizomorph morphology, stipe-scale colour,

poor development of crassobasidia in the African material and lack of clamp-connections in the Sri Lankan material. However, later, after Watling had examined material on which A. heimii Pegler and Clitocybe elegans were each based (in Kile & Watling, 1988) Pegler's opinion based on morphological characteristics was supported; cultural and interfertility studies are necessary to confirm this view and are now under way (Mohamed, pers. comm.).

Petch (1928) draws a similarity between A. fuscipes and illustrations of A. mellea from the Gold Coast (Bunting & Dade, 1924) on cocoa (Theobroma cacao L. - Sterculiaceae) and Java (Bernard & Palm, 1918) where it causes a split root disease of tea (Camellia sinensis (L.) Kuntze - Theaceae). Undoubtedly Petch's decision that his and Bunting & Dade's fungi are the same is correct and they agree with Heim's fungus Clitocybe elegans. Indeed Petch's black and white photograph taken of material matured in the laboratory in Peradeniya (Petch, 1923) could have been of the Korup fungus.

Armillaria fuscipes, as A. heimii, has been recorded by Pegler (1977) on old dead stumps in natural forest in Tanzania, and on rotting stumps in the Mpanga For. Reserve, Uganda. On introducing the name, Pegler (1977) also associated the fungus with tea plantations in Tanzania. Ivory has kindly supplied material (Wat. 19722 in E) isolated from Tectona grandis L.f. (Verbenaceae) in Zambia quite close to a site from which Pegler had identified his A. heimii (Ivory, pers. comm.).

Heim (1967) records A. fuscipes (as Armillariella elegans) from the Western Highlands of New Guinea and this highlights the similarities between A. fellea and A. fuscipes (Kile & Watling, 1988) and Bernard & Palm's record from Java above. It would appear that more observations are required on the S.E. Asian members of the genus to complete our understanding of the members of the genus Armillaria in the arc of countries adjacent to the Indian Ocean.

# NOMENCLATURAL SUMMARY

Armillaria fuscipes Petch in Ann. bot. Gdns Peradeniya 4: 299. 1909.

Clitocybe (Armillariella) elegans Heim in Revue Mycol. 28: 94. 1963; non Armillaria elegans Beeli in Bull. Soc. r. Bot. Belg. 109: 111. 1927 (= Cystoderma). — Armillariella elegans (Heim) Heim in Revue Mycol. 32: 9. 1967 (not validly published).

Armillaria heimii Pegler in Kew Bull. addit. Ser. 6: 92, 1977 (name change for C. elegans Heim).

Excluded.—Armillaria mellea var. camerunensis Henn. in Bot. Jb. 22: 107. 1895. — Armillariella camerunensis (Henn.) Singer, Agaricales modern. taxonomy (ed. 4) 263. 1986.

### CONCLUSION

It would appear that the correct name for Clitocybe elegans is Armillaria fuscipes originally described from Ceylon (Sri Lanka). It was very probably introduced there when the coffee industry collapsed because of the ravages of the rust-fungus Hemileia vastatrix Berk. & Broome in the early part of the century and the plantations were replaced by the then expanding tea industry. A possible source of inoculum was from this same industry in East Africa where there was an interchange of materials and personnel. It would appear that A. fuscipes occurs naturally in the rainforest of West Central Africa and Madagascar where it does not appear to

be causing extensive death of native trees; in the Cameroon the area in which it has been collected is thought to be a Pleistocene refugia. Wherever plantations of cocoa, coffee, tea or Albizzia have been introduced, widespread trouble from this fungus is recorded reflecting a parallel phenomenon to that in northern British Isles where A. borealis Korhonen, normally causing little damage in birch woods, takes on a primary parasitic role in plantation trees when the birch is replaced by conifers in upland sites. Armillaria luteo-bubalina Kile & Watling develops a similar strategy in Australia in citrus and garden plantings after the clearing of native dry sclerophyll scrub, a habitat in which A. luteo-bubalina is native and widespread.

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